

## ***Laboratory Clinical Specimen Collection and Storage Guidance For Lung Injury Related to e-Cigarette Related Exposures***

As this investigation continues, CDC encourages clinicians to report possible cases of e-cigarette or vaping-associated pulmonary disease to their [local or state health department](#) for further investigation.

If e-cigarette or vaping product use is suspected as a possible cause for a patient's pulmonary disease, a detailed history of the substances used, the sources, and the devices used should be obtained, as outlined in [the Health Alert Network \(HAN\)](#), and efforts should be made to determine if any remaining product, devices, and liquids are available for testing.

While testing of vaping liquids and devices is ongoing CDC has been asked to provide guidance on collection and storage of clinical specimens. This may include retention of residual clinical specimens collected for patient care or specimens collected and stored from early in a patient's presentation to look for markers of exposure.

Decisions about the collection and storage of clinical samples should be made in collaboration between healthcare providers and state public health authorities. Early communication should be initiated with your laboratory to retain samples if further analysis is anticipated. This guidance will be updated based on the ongoing investigation.

### **Scope**

The purpose of this document is to provide **general specimen collection and storage guidance** for healthcare providers and public health laboratory personnel involved in the care of patients who meet, or are highly suspected of meeting, the case definition for severe pulmonary disease related to e-cigarette use or vaping. These recommendations are not intended to direct laboratory guidance for patient care. Empiric collection instructions are limited to collection and storage of blood and urine. For the patients who have undergone bronchoalveolar lavage (BAL) instructions are provided for preservation of BAL fluid.

**NOTE: The decision to collect samples is at the discretion of healthcare providers and to store samples is at the discretion of healthcare providers and state public health authorities.**

To date, there are no identified chemical or biological markers that would warrant specific recommendations for analysis of blood or urine.

This guidance is provided as a proposed best practice for timely specimen collection, handling, and retention in case a useful marker specific to respiratory disease caused by e-cigarette products is identified in the future.

Retention and storage of residual specimens that were collected for other types of diagnostic screening and testing can also be considered. Healthcare providers should contact their hospital laboratories to identify and retain such specimens before disposal.

Long term storage should be coordinated with the state public health authorities and state public health laboratories.

**NOTE: CDC also recommends consulting your local or state health department about the recommendations on collection and storage of samples – in addition to this guidance.**

### **Specimen Collection – for Healthcare Providers and Clinical Laboratory Personnel**

## ***Laboratory Clinical Specimen Collection and Storage Guidance For Lung Injury Related to e-Cigarette Related Exposures***

### **Blood Samples**

1. For each patient, collect up to 8 mL of blood in two (2) 4-mL **PURPLE**-top (K2 -EDTA) glass or plastic tubes. If only 3-mL tubes are available, three (3) 3-mL tubes may be collected. (Note: **DO NOT** use gel separators.)
2. Mix contents of tubes by inverting them 5 or 6 times.
3. Label tubes in order of collection. Example: #1, #2, #3.
4. Place a barcode label on each tube so that the barcode looks like a ladder when the tube is upright.
5. Store blood samples at 1 °C to 10 °C. **DO NOT FREEZE** if prompt transfer to state lab is anticipated
6. If transfer to state lab is expected to be longer than 6 hours. Separate plasma from whole blood cells within 6 hours of collection. Aliquot plasma into cryotubes

### **Urine Samples**

1. For each patient, store 40 to 60 mL of urine in a screw-cap urine cup.
2. Place barcoded label on the cup when upright; the barcode will look like a ladder.
3. Indicate on the cup how the sample was collected if the method was other than “clean catch” (Example: catheterization).
4. **Store urine samples in the freezer.** Freezer temperatures of -20 °C or lower are recommended.

Following the above collection protocol, contact your state public health laboratory for specimen transport and long-term storage.

## ***Laboratory Clinical Specimen Collection and Storage Guidance For Lung Injury Related to e-Cigarette Related Exposures***

### **Bronchoalveolar Lavage Fluid (BAL fluid)**

1. Due to the increased technical skill and equipment needed and invasive nature of sampling the decision and timing for undergoing BAL should be left to the treating clinicians.

2. **Optimal timing.** These specimens may be obtained at any time during the clinical course, but ideally prior to initiation of antimicrobial or steroid therapy. If antibiotics or steroids have been initiated course and duration should be noted.

### **3. Specimen collection.**

#### **BAL fluid**

Collect specimens in sterile containers.

BAL fluid should undergo culture and routine centrifugation followed by cellular analyses and cytopathology, including lipid and other staining, as clinically indicated at the local institution.

Lipoid pneumonia associated with inhalation of lipids in aerosols generated by e-cigarettes has been reported based on the detection of lipid-laden alveolar macrophages obtained by bronchoalveolar lavage (BAL) and lipid staining (e.g., oil red O).

**This guidance is primarily to retain sample leftover from routine clinical evaluation.**

Remaining uncentrifuged BAL fluid and supernatant from centrifuged BAL fluid should be labeled as such and be retained. Up to 10 unstained cytology slides should be briefly fixed in formalin and retained for future evaluation.

Excess cell pellet after cytopathologic evaluation can be divided in half with half being fixed in formalin and stored at room temperature for further cytopathologic evaluation and half frozen at -20 °C or lower for future chemical or lipid analysis.

Place remaining uncentrifuged fluid and centrifuged supernatant from centrifuged fluid into sterile vials with external caps and internal O-ring seals. If there is no internal O-ring seal, then seal tightly with the available cap and secure with Parafilm®. Label each specimen container with the patient's name, ID number, the specimen type, subsection of lung lavaged and the date the specimen was collected.

### **Long-term Storage – for Public Health Laboratory Personnel**

1. Establish communication protocols with local hospitals and other medical providers regarding appropriate specimen collection guidance (see above).
2. Plan to transport collected specimens on at least a bi-weekly (or more frequently if needed) basis.
3. Follow your local in-house chain of custody protocols (i.e., shipping manifests).
4. Collect specimens from local health providers.
5. Separate plasma from whole blood cells within 6 hours of collection. Aliquot plasma into cryotubes
6. **FREEZE** specimens (urine, plasma, BAL fluid) at freezer temperatures of -20 °C or lower.
7. Any formalin-fixed specimens (BAL pellet, cytology slides) should be stored at **room temperature NOT FROZEN**.

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8. Contact your state epidemiologist or outbreak principal investigator regarding next steps.

**Note:** Consult your local or state health department about the outbreak of lung disease associated with e-cigarette use (vaping) as soon as possible. State health department officials seeking technical assistance with case reporting or epidemiologic issues can contact CDC at [VapingAssocIllness@cdc.gov](mailto:VapingAssocIllness@cdc.gov). State health department officials seeking technical assistance with laboratory testing can discuss with their state health department laboratories or contact CDC at [VapingAssocIllness@cdc.gov](mailto:VapingAssocIllness@cdc.gov).